

ON THE ANTAGONISM BETWEEN PILOCARPINE
AND EXTRACT OF AMANITA MUSCARIA.

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WE are anxious at the outset to acknowledge our indebtedness to Mr E. A. Morshead, who largely assisted us in the experiments performed in July.

Pituri, as we have shown¹, excites salivation and perspiration, corresponding in this respect to muscarin and pilocarpine. Pilocarpine and muscarin, however slow, weaken and even arrest the heart, whilst, if we may assume this from one experiment, pituri, topically applied, has little or no action on the cardiac contractions. Pituri moreover antagonizes the action of muscarin and pilocarpine on the heart; thus we have a sweater and a salivator antagonizing the action on the heart of substances which likewise cause salivation and perspiration.

It occurred to us that possibly muscarin and pilocarpine, though they affect the heart in a similar manner, might antagonize each other's effects, and we now record some experiments designed to test the validity of this conjecture.

In all these experiments we first killed the frog by dividing the spinal cord opposite the occipito-atlantoid membrane, and destroying the brain by thrusting a peg into the skull. We then, without dividing the sternum, exposed the heart and cut through the pericardium.

We used an alcoholic extract of amanita muscaria, and a solution of nitrate of pilocarpine, in some cases a two per cent., in others a ten per cent. solution.

Jan. 3. To a heart beating 44 per minute we applied a small quantity of extract of muscaria. In four minutes the heart stopped, widely distended with blood. Three quarters of a minute afterwards we applied some 10 p. c.

¹ *Journal of Phys.*, 1878, p. 377, and *Lancet*, 1879.

solution of nitrate of pilocarpine, when the heart almost immediately began to contract, and in one minute it beat well at 12 per minute, and in forty-five minutes it beat 24. The strength of the contractions then again grew weaker, till in thirty-eight minutes it beat very feebly at 20. Then we applied a little atropia, and at once the beats improved much in strength, rose to 24, and in twenty-two minutes the beats were good at 40 per minute, and six hours later the heart still contracted well at 36 per minute.

On the same day we repeated the experiment. To the exposed heart we applied a small quantity of muscarin, and in eight minutes the pulsations fell from 40 to between 4 and 6 per minute. We then applied a little of the 10 p. e. solution of nitrate of pilocarpine, and in half a minute the pulsations rose to 28 very good beats; in 13 minutes the pulsations began to grow feeble, and in 44 minutes dropped to 12 per minute, and were so feeble as to be scarcely visible. Five hours afterwards the heart was motionless, flaccid, and contained a little blood, having we felt no doubt long ceased to beat, since general reflex action had quite ceased; still we determined to apply a little of the 1 p. c. solution of nitrate of pituria, not for a moment expecting that it could restore the lost cardiac action: but to our astonishment in six minutes the heart began to contract; in ten minutes it beat faintly at 16 per minute; in twelve minutes the contractions were much stronger; in fourteen minutes it beat fairly well at 18, and an hour and a quarter at 20 per minute.

Last summer one of us, in conjunction with Mr E. A. Morshead, made a series of observations regarding the antagonism of pilocarpine for muscarin, making use of a solution of muscarin containing one part of alcoholic extract of *amanita muscaria* in four parts of water, and one part of nitrate of pilocarpine in twelve parts of water.

July 10. Whilst the heart beat at 54 we applied the muscarin solution freely, which at once slowed the heart to 46, and gradually in the course of thirteen minutes the beats fell to 32 and grew feebler, so that at the expiration of this time the heart contracted hardly at all and remained widely distended. Then we applied some of the pilocarpine solution, when the contractions at once very much improved and became fairly strong, with unaltered frequency for ten minutes, and then rose to 44. The heart continued to beat fairly well at 44 for five hours; in ten hours afterwards it beat feebly at 44, and in twenty-four hours it had stopped, firmly contracted.

July 10. Muscarin solution applied to the heart beating at 58 at once weakened the contractions and reduced them to 34, and this number gradually fell till in eleven minutes it had dropped to 22, when the auricles had ceased to contract and the ventricle contracted very slightly indeed. At this stage we freely applied the pilocarpine solution, and after a pause of some seconds the ventricle at once commenced work fairly well at 28, and in three minutes both auricles and ventricle contracted firmly at 34; in fact as strongly as before the application of the muscarin, and so continued during nine minutes. On our return in two hours we found the heart stopped in wide diastole.

July 10. To a heart beating at 46 we applied some muscarin solution and the pulsation fell in two minutes to 28 and almost stopped, the ventricle, widely distended with blood, alone contracting in the slightest degree, so that only a slight quiver of its walls could be seen. We then applied pilocarpine. In two minutes the ventricle contracted strongly; in six both auricles and ventricle contracted well at 30; the contractions then rose to 44; ten hours later the heart beat well at 44, and even twenty-four hours later it still contracted slightly.

July 11. To a heart beating at 54 we applied the muscarin solution, and in six minutes the pulsation ceased. Then we applied some pilocarpine solution. In one minute pulsation recommenced, and in two minutes rose to 24. In seven minutes we reapplied the pilocarpine, and in thirteen minutes from the second application the heart beat well at 24; the pulsation then grew slower and feebler, so in fifty-three minutes after the first application we again applied pilocarpine, but without strengthening the heart, which continued to beat at 12 per minute. We then reapplied some muscarin and subsequently some more pilocarpine, but the heart never improved, and in fifty-eight minutes from the first application of the pilocarpine only the auricle beat very faintly.

July 11. To the heart beating at 44 we applied some muscarin solution. The pulsations fell to 30 and the auricle stopped, whilst so feeble was the contraction of the ventricle that apparently it was unable to propel any blood. Then we applied pilocarpine and the pulsations rose to 34, and both auricle and ventricle contracted fairly well. In twenty-seven minutes the contractions became feebler, and on returning two hours after the heart had stopped, widely distended with blood.

July 11. To a heart beating at 46 we applied some muscarin. In four minutes the heart stopped, widely distended with blood. We then applied some pilocarpine solution, and for the next six minutes no contractions occurred; in nine minutes the auricles contracted slightly; in fourteen minutes we reapplied pilocarpine, still only the auricles contracted, and fairly well at 32.

It occurred to us that possibly the revivifying effect of pilocarpine applied to the heart previously weakened or arrested by muscarin might be simply a physical result caused by the application of the solution, and not a specific effect of the pilocarpine, and we resolved to make an experiment to ascertain whether this might be a correct surmise.

On Jan. 3 we applied a little muscarin to a brainless frog's heart beating well at 32 per minute. During the ensuing five minutes the beats fell to 4, but were fairly good. We then washed the heart well for five minutes with a 1 p. e. saline solution, but without in the slightest degree quickening or strengthening its beats. Then we applied a little (1 in 10) pilocarpine solution. In two minutes the heart grew much stronger and beat 16, but in four minutes it again grew weaker and fell to 8. Eight minutes later we again applied pilocarpine, and in six minutes the heart, pulsating well, rose to 16. It continued in the same condition thirty-two minutes after the second application of pilocarpine and then grew weaker,

though still it continued beating at 16. We then washed the heart well for three minutes with saline solution without affecting either its force or frequency, and an hour after the second pilocarpine application, with the heart at 16, we added a little 1 p. c. solution of sulphate of atropia, when the pulsations improved and rose to 24.

This experiment confirms the preceding, and shows that pilocarpine antagonizes for a short time the effect of muscarin, and that this revival of the heart's action is not due to the mechanical effect of the pilocarpine solution, for a saline solution applied freely for five minutes produced no effect.

Table I.

Date.	After the application of muscarin the pulsations fell	After the application of muscarin the pulsations became	After the application of pilocarpine the pulsations rose	After the application of pilocarpine the pulsations became	The pulsations continued	Pulsations had ceased in
Jan. 3	stopped	stopped	24 beats	good		
„ 3	36 beats	„	24 beats	good		5 hours
„ 3	28 beats	remained fairly good	12 beats	much stronger		
July 10	22 beats	almost stopped	12 beats	much stronger	10 hours	
„ 10	36 beats	ventricle alone contracted and very feebly	12 beats	good		2 hours
„ 10	18 beats	almost stopped, auricle quite stopped	16 beats	good	24 hours	
„ 11	54 beats	stopped	24 beats	good		1 hour
„ 11	14 beats	auricle stopped and ventricle contracted very feebly	4 beats	fairly good		2 hours
„ 11	stopped	stopped	auricles only at 32	feeble and only in auricle		1 hour

A glance at Table I., in which we have summarised the results of the foregoing experiments, shows at once that pilocarpine in a great measure antagonizes the action of muscarin on the heart; that pilocarpine restores vigorous contractions to an arrested heart and strengthens and increases the frequency of a slowed and much weakened heart.

The table further shows that the antagonistic action of pilocarpine is displayed more obviously in increasing the strength than the frequency of the contractions.

Here we have another remarkable instance of an antagonistic action between two drugs otherwise so similar in their effect on the animal body: an instance, too, of antagonistic action in an organ though their effect when separately applied is similar; for both muscarin and pilocarpine slow and weaken or even arrest the heart, and yet, as we have just seen, pilocarpine antagonizes the action of muscarin.

It may possibly be said that the improvement from the application of the pilocarpine is a mere coincidence, and would occur irrespective of it; but to this we answer that in all our experiments prior to the application of the pilocarpine no improvement took place, whilst in every instance more or less amendment occurred immediately after the pilocarpine. Still we think it well to give experimental evidence on this point.

We therefore resolved to perform a set of comparative test experiments:

- i. The action of muscarin on the heart.
- ii. Of muscarin followed by pilocarpine.
- iii. Of muscarin followed by atropia.
- iv. Of muscarin followed by duboisia.

We destroyed the brains of three frogs and then laid bare their hearts. To the first we applied muscarin only. To the second we applied first muscarin, and when this had affected the heart we applied pilocarpine. To the third we applied first muscarin, and when the heart became fully affected we applied atropia. By means of these comparative experiments we can ascertain the degree of antagonism which pilocarpine and atropia manifest to muscarin, and we are able to compare the degree of antagonism exerted by pilocarpine with that exerted by atropia.

We used a 2 p. c. solution of nitrate of pilocarpine, a 1 in 120 solution of sulphate of atropia, and a solution of muscarin containing one part of alcoholic extract of *amanita muscaria* in four of water, and a 1 in 120 solution of the alkaloid of duboisia. The pilocarpine and extract of *ammonita* solution has a slight acid reaction. The results are given in Tables II. to V.

The experiments given in Table II. fully confirm the previous ones. They show that the heart does not recover after it is slowed or arrested by muscarin, and consequently that the improvement following the application of pilocarpine is not a natural recovery that would have occurred independently of the pilocarpine, but is due to the action of that drug. We now summarize the results given in Tables I. III. IV. and V.

In the first experiments given in Table I. extract of *muscaria* stopped the heart in four cases and almost stopped it in the remainder,

whilst pilocarpine increased the pulsations on the average 18 beats, induced good cardiac contractions in seven cases, fairly good in one, and restored merely slight movement in the auricle in one case after arrest of the heart.

In the six experiments given in Table III. the muscaria stopped the heart in two cases and almost stopped it in the rest, whilst the pilocarpine increased the pulsations 16 beats on the average, and in three instances induced very good pulsations, in one good, and in two fairly good.

Our second series of experiments, Table III., confirms the first, Table I., in showing that, in a heart weakened and slowed by extract of muscaria, pilocarpine restores the strength even more than the frequency of the beats.

Our second series of observations enables us to compare the antagonistic action of pilocarpine with that of atropia and duboisia. The average increase in the number of beats after atropia and duboisia is as follows: after atropia 28, after duboisia 30.

Both in the experiments with atropia and duboisia the heart in every instance was stopped or almost stopped, and again on the application of either atropia or duboisia the drug in every case induced good pulsations.

The antagonism of pilocarpine for extract of amanita muscaria is not so marked as the antagonism of atropia or duboisia for muscarin. Atropia and duboisia increasing the pulsation of a heart stopped by muscarin on the average about 30 beats per minute, whilst pilocarpine augments the beat only to about 16 to 18 per minute; but the increase in the strength of the beat was as great after the application of pilocarpine as after the application of atropia or duboisia.

The greater antagonizing power of atropia and duboisia over pilocarpine is shown by the longer time the heart continues to beat after the application of atropia or duboisia, thus after

pilocarpine	the heart stopped on the average in 28 hours
atropia 47 „
duboisia 49 „

We next adopted another method of comparing the antagonism of pilocarpine with the antagonism of atropia for muscarin.

We first arrested the heart with extract of amanita muscaria, and then allowed 8, 15, and 25 minutes to elapse before we applied either the pilocarpine or the atropia, using solutions of the same strength as in the previous experiments. The results we give in Tables VI. and VII.

Table II.
Action of muscarin on the Heart.
The heart beating at 40, fell in a $\frac{1}{4}$ of an hour to 4, and stopped in an hour and did not begin again.

Jan. 10	44	5 minutes	5	25 minutes	"
" 14	40	7	4	12	"
" 15	36	9	4	28	"
" 21	36	7	4	23	"
" 22	36	8	12	20	"
" 24					"

Table III.
Antagonism of pilocarpine to muscarin.

Date.	After application of muscarin pulsations fell	After application of muscarin heart became	After application of pilocarpine rose pulsations	After application of pilocarpine heart became	Pulsation continued
Jan. 10	in 2 min. from 40 to 4	very feeble, all but stopped	in 17 min. to 20	very good	2 hours after became very weak; 12 hours after beat feebly at 4 per min.
" 14	in 15 " 40 to 4	" "	in 17 min. to 10, and in 44 min. to 12	fairly good	stopped in 4 hours
" 15	in 7 " 40 to 4	" "	in 6 min. to 10, and in 12 hours to 20	very good	15 hrs. after good at 20; 40 hrs. after at 16; stopped in 48 hrs.
" 21	in 9 " 40 to 2 stopped in 28 min.	stopped	in 9 min. from 0 to 20	good	36 hrs. after good at 20; 60 hrs. after good at 10; stopped 66 hrs.
" 22	in 7 min. from 22 to 4 stopped in 13 min.	stopped	in 14 " 0 to 20	very good	24 hrs. after good at 16; stopped 31 hrs. after
" 24	in 2 min. from 38 to 4	almost stopped	in 2 " 4 to 12 in 15 min. to 20	fair	stopped in 7 hrs.

Table IV.
Antagonism of atropia to muscarin.

Date.	After application of muscarin pulsation fell	Pulsations became	After application of atropia the pulsation rose	The pulsations became	Pulsations continued
Jan. 10	in 2 min. from 40 to 4	very feeble	in 2 min. from 4 to 32	very good	12 hours after pulsation very good at 30, and 84 hours after beat fairly well at 36
" 14	in 13 " 60 to 6	very feeble	in 1 " 6 to 40 & in 30 min. rose to 52	"	36 hours after beat well at 36, and 48 hours after very feebly
" 15	in 12 " 52 to 0	stopped	in 1 min. 0 to 34	"	50 hours after beat well at 36
" 21	in 4 " 40 to 4	stopped	in 2 " 0 to 20	good	10 hours after had stopped
" 22	stopped in 28 min. in 3 min. from 28 to 4	very feeble	in 3 " 4 to 24	"	36 hours after beat was at 26; 44 hours afterwards stopped
" 24	in 4 " 28 to 2	almost stopped	in 1 " 2 to 24	"	12 hours after good at 24

Table V.
Antagonism of duboisia to muscarin.

Jan. 14	in 5 min. from 48 to 0	arrested for 3 min.	in 3 min. from 0 to 40	very good	15 hours after beat fell at 24;
" 15	in 7 " 52 to 0	stopped	in 3 " 0 to 28	good	stopped in 36 hours
" 21	in 7 " 40 to 4	stopped	in 3 " 0 to 36	very good	36 hours after good at 24; 59 hours beat feebly at 16; stopped in 56 hours
" 22	in 3 min. from 24 to 8 and in 27 min. to 4	very feeble	in 3 " 4 to 20 and in 2 hrs. rose to 24	"	63 hours after good at 24
" 24	in 9 min. from 36 to 4	almost stopped	in 1 min. rose to 30	good	12 hours after good at 28

Table VI.

Date.	After the application of extract of amanita muscaria pulsations stopped in	After the application of pilocarpine the pulsations begun in	and rose to	and became	Pulsations before application of muscarin.
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An interval of eight minutes after arrest by muscarin.

Feb. 26	29 min.	2 min.	24 in 44 min.	good at once	40.
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An interval of fifteen minutes after arrest by muscarin.

Feb. 26	65 min.	4 min.	20 in 20 min.	very good at once	38.
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An interval of twenty-five minutes after arrest by muscarin.

Feb. 27	at once	not at all			40. The heart was watched for 4 hrs.
Feb. 28	3 min.	„			24 to 28.
Mar. 3	48 min.	34 min.*	20 in 34 min. to 32 in 55 m.	very good	56. 14 hours afterwards the ventricle beat poorly at 8.

* The auricle begun to beat 8 minutes after the application of pilocarpine. In our figures in these tables we refer to the ventricle.

Table VII.

Date.	After the application of extract of amanita muscaria pulsations ceased in	After the application of atropia the pulsations begun in	and rose to	and became	Pulsations before application of muscarin.
<i>An interval of eight minutes after arrest by muscarin.</i>					
Feb. 26	20 min.	3 min.	32 in 75 min.	good at once	40.
<i>An interval of fifteen minutes after arrest by muscarin.</i>					
Feb. 26	29 min.	7 min.	12 to 20 in 7 min.	fairly good	40.
<i>An interval of twenty-five minutes after arrest by muscarin.</i>					
Feb. 27	at once	11 min.	20 in 19 min.	good	48.
Feb. 28	14 min.	10 min.	14 in 16 min.	good	56. 14 hours afterwards beat at 8, but had become weaker.
Mar. 3	17 min.	17 min.	28 in 100 min. 36 in 14 hrs.	very good	56.

Our experiments then show that pilocarpine, a sweater and salivator which slows and weakens or arrests the frog's heart, antagonizes, as regards the heart, the action of extract of amanita muscaria (muscarin), a substance which is also a sweater and salivator, and slows and also weakens or arrests the frog's heart.

In this respect pilocarpine is but little inferior to atropia and du-boisia.